ABSTRACT

The invention comprises methods of detecting the presence or level of an analyte in a sample by detecting the formation of a binding complex on a solid phase. Preferred "competition-like" methods of the invention comprise the steps of:

(a) mixing the sample with a second ligand capable of binding with said analyte so that an analyte/second ligand complex is formed;

(b) contacting the mixture produced in step (a) with a solid phase having bound thereto a first ligand capable of binding with the second ligand so that a first ligand/second ligand complex is formed, the contacting being performed under conditions and for a time sufficiently limited that dissociation of the analyte/second ligand complex formed in step (a) is substantially inhibited;

(c) binding a detectable tag to the second ligand either prior to or after step (a) or step (b) so that a portion of the tag is retained on the solid phase upon formation of the first ligand/second ligand complex;

(d) detecting the portion of the tag to detect formation of the first ligand/second ligand complex on the solid phase, so that the presence or level of the analyte in the sample can be determined.

Preferred "sandwich-type" methods of the invention comprise the steps of:

(a) contacting the sample with:

(i) a solid phase having bound thereto a first ligand capable of binding the analyte; and

(ii) a second ligand capable of binding to the first ligand or to a first ligand/analyte complex so that a first ligand/analyte/second ligand complex is formed on the solid phase, the contacting being performed under conditions and for a time sufficiently limited that any non-specific binding between the second ligand and the solid phase is substantially inhibited;

(b) binding a detectable tag to the second ligand either prior to or after formation of the first ligand/analyte/second ligand complex so that a portion of the tag is retained on the solid phase upon formation of the first ligand/analyte/second ligand complex;

(c) detecting the tag to determine the presence or level of the analyte in the sample.

The invention also provides methods for single-point calibration and quality assurance that can be used in conjunction with the above-mentioned competition-like and/or sandwich-type assays.

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